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Notes on Histological Technique.*

MOUNTING OF ALGAE.

J. A. NIEUWLAND.

In a personal communication from the well-known phycologist Professor G. S. West, of the University of Birmingham, England, I received an outline of a method of fixing, mounting, and preserving algae which, as he tells me, has not been given the attention that it perhaps deserves. The fluid used serves at the same time as a killing, fixing, preserving, and mounting medium, and for delicate structures like desmids and other algal forms, it is said not to be surpassed. It has the advantage, moreover, that it keeps the natural coloring of the green algae, something which the instructor in elementary laboratory work will appreciate better than anyone else. The fluid is a 2 per cent. solution of potassium acetate, just made blue with a small amount of copper acetate. The substance reduces plasmolysis of the cell contents to a minimum. The algae can be put into the solution and kept in it. If a permanent mount is wanted a small amount of the material is put on a rather thick slide and sealed with old gold-size several times after each drying. The mounts are permanent, but it is necessary to take great care in sealing, and to this end to use a thick slide. A thin slide will bend considerably in handling, and the sealing may be separated in this way from the slide, so that the preparation will dry up as the result.

For some reason the fluid presents considerable difficulty with *Vaucheria*, and plasmolysis is hard to avoid. I have found that the best way to treat *Vaucheria*, especially the zoospores before or just after germination, when the plant is particularly delicate, is to kill it rapidly with 3 or 4 per cent. formalin. The formalin must be completely and quickly removed or the preparation will turn black afterward. Fixing for half an hour in the 3 per cent. formalin will not be injurious. Remove the formalin by repeated washing with water. If the *Vaucheria* thus treated is rapidly brought into glycerin to which a little thymol is added, the preparation will be

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as perfectly green as when alive, and will retain its green color indefinitely. The method may be extended to all small green forms like the smaller liverworts, fern prothallia, and moss protonemata.

To get the material into glycerin, add first a considerable quantity of 5 or 10 per cent. glycerin in water, and put the dish near but not on a radiator. In a few days the evaporation will leave the fluid thick. Once the preparation is in thick glycerin the color will not change, if the formalin has been completely removed.

The potassium-copper-acetate solution will not keep the natural color of diatoms. It has the property of removing the diatomin or yellow coloring matter from the diatoms and leaving the plants perfectly green. The solution can thus be used in demonstrating the presence of chlorophyll in these plants. The diatomin is removed or absorbed in a few minutes after application.

As I have found some difficulty in keeping the microscopic mounts made in the potassium-copper-acetate solution because of drying, I have evolved a modification of the glycerin method in combination with it. The mounts made by this method are perfectly durable, and when carefully prepared are superior to ordinary glycerin mounts, as all the green algae treated with it keep their natural colors indefinitely. Glycerin jelly can also be used at the end to make the mount even more durable than the ordinary glycerin mount would be. The procedure is as follows.

The algae to be used are fixed in the potassium-copper-acetate 2 per cent. solution. After they have been killed and fixed in this fluid (the time varying according to the specimen treated), add to the above solution an equal part of 10 per cent. glycerin solution and allow to concentrate by evaporation in a warm dry place protected from dust. The algae must be thoroughly separated from dirt and soil or the concentrated solution will precipitate a reddish-brown cloud of reduced copper. In nearly all cases the preparation when thickened will be covered with a film of acetates, which can be removed from the top of the fluid without injury to the material. The concentrated solution should be perfectly clear, of a light green color, and the chromatophores of the algae perfectly green as in life. I have often been asked by students, and in fact by those well acquainted with algae, whether the plants thus given them for examination were not really alive. The advantage of having plant material, especially for elementary students, in a condition as near as possible to the live state, obviates explanations about stains. I

have found it very undesirable to give beginners any material other than alive or such as looks like the live stage of the plant studied.

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Teratological Notes.

II. AN ABNORMAL FLOWER OF *CAMPANULA ROTUNDIFOLIA*.

A somewhat similar case of abnormal growth as was noted in the first number of the *Midland Naturalist* by Professor Kirsch in the case of *Taraxacum officinale*, Weber, was observed during the summer of 1906, in *Campanula rotundifolia*, L. This peculiar specimen of the Common Bluebell was found at Notre Dame, Ind., north of St. Joseph's Lake, on a dry sunny slope of a hill. The extraordinary shape and size of the terminal flower of the plant made it an object that caught immediate attention. The whole flowering axis of the plant was picked and is now preserved in four per cent. formalin.

The main stem and the peduncle of the plant from the root leaves to the abnormal flower at the tip, is flattened throughout and about one-fourth of an inch in width. Branching from this main axis appear at intervals a number of pedicels with normal flowers with five parts to their calyx and corolla. The apical abnormal flower is in general somewhat flattened. Its calyx is synsepalous and attached to the ovary the usual height as in ordinary flowers, but it has 32 teeth instead of the usual 5. The calyx-teeth are similar to those of an ordinary flower and all of equal size and perfectly formed, and 28 lobes of the corolla are present, these also of normal appearance. On account of the large number of the lobes of the sympetalous corolla, the flower has rather the appearance of a cow-bell, than an ordinary bell with the parts crimped and doubled in upon themselves in several places. The flower is perfectly full-blown and the anthers have in large part fallen away, adding to the difficulty of finding the exact number at present. The stamens must have been about 27-30. The style of the flower is flattened with the upper stigmatic part thickened, the